

VCPIP1 siRNA (h): sc-77549

BACKGROUND

VCPIP1 (valosin containing protein (p97)/p47 complex interacting protein 1), also known as DUBA3 or VCIP135, is a 1,222 amino acid protein that contains one OTU domain and localizes to the endoplasmic reticulum (ER), as well as to Golgi stacks within the Golgi apparatus. Interacting with VCP (valosin-containing protein) and p47, VCPIP1 functions as a deubiquitinating enzyme that is necessary for post-mitotic golgi stack formation and may also play a role in the VCP-mediated creation of the transitional ER (tER). The gene encoding VCPIP1 maps to human chromosome 8, which consists of nearly 146 million base pairs, houses more than 800 genes and is associated with a variety of diseases and malignancies. Schizophrenia, bipolar disorder, Trisomy 8, Pfeiffer syndrome, congenital hypothyroidism, Waardenburg syndrome and some leukemias and lymphomas are thought to occur as a result of defects in specific genes that maps to chromosome 8.

REFERENCES

1. Uchiyama, K., et al. 2002. VCIP135, a novel essential factor for p97/p47-mediated membrane fusion, is required for Golgi and ER assembly *in vivo*. *J. Cell Biol.* 159: 855-866.
2. Ficarro, S., et al. 2003. Phosphoproteome analysis of capacitated human sperm. Evidence of tyrosine phosphorylation of a kinase-anchoring protein 3 and valosin-containing protein/p97 during capacitation. *J. Biol. Chem.* 278: 11579-11589.
3. Wang, Y., et al. 2004. VCIP135 acts as a deubiquitinating enzyme during p97-p47-mediated reassembly of mitotic Golgi fragments. *J. Cell Biol.* 164: 973-978.
4. Kano, F., et al. 2005. NSF/SNAPs and p97/p47/VCIP135 are sequentially required for cell cycle-dependent reformation of the ER network. *Genes Cells* 10: 989-999.
5. Uchiyama, K., et al. 2005. p97/p47-mediated biogenesis of Golgi and ER. *J. Biochem.* 137: 115-119.
6. Uchiyama, K., et al. 2006. p37 is a p97 adaptor required for Golgi and ER biogenesis in interphase and at the end of mitosis. *Dev. Cell* 11: 803-816.

CHROMOSOMAL LOCATION

Genetic locus: VCPIP1 (human) mapping to 8q13.1.

PRODUCT

VCPIP1 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see VCPIP1 shRNA Plasmid (h): sc-77549-SH and VCPIP1 shRNA (h) Lentiviral Particles: sc-77549-V as alternate gene silencing products.

For independent verification of VCPIP1 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-77549A, sc-77549B and sc-77549C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

VCPIP1 siRNA (h) is recommended for the inhibition of VCPIP1 expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

VCPIP1 (C-12): sc-515291 is recommended as a control antibody for monitoring of VCPIP1 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker[™] Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz[®] Mounting Medium: sc-24941 or UltraCruz[®] Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor VCPIP1 gene expression knockdown using RT-PCR Primer: VCPIP1 (h)-PR: sc-77549-PR (20 μ l, 592 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.